Global Veterinaria 8 (4): 393-396, 2012
ISSN 1992-6197
© IDOSI Publications, 2012

Study of the Combination Effects of Aflatoxin and T-2 Toxin on Performance Parameters and Internal Organs of Commercial Broilers

[1]M. Manafi, [2]B. Umakantha, [3]M. Noor Ali and [4]H.D. Narayana Swamy

[1]Department of Animal Science, Malayer University, Malayer, Iran
[2]Department of Poultry Science, Veterinary College, KVAFSU, Bangalore, India
[3]Faculty of Veterinary Science, Herat University, Herat, Afghanistan
[4]Department of Veterinary Pathology, Veterinary College, KVAFSU, Bangalore, India

Abstract: The additive effects of aflatoxin (AF) and T-2 toxin (T-2) on the performance of broiler chickens were evaluated, individually and in combination. One hundred and sixty eight day-old Cobb broiler chicks were obtained from a commercial hatchery and divided into four groups in 2X2 Complete Randomized Design of three replicates and fourteen chicks per replicate, with dietary treatments of 0.0 (control), 0.5µg/g AF, 2.0µg/g T-2 and their combination (0.5 µg/g AF+2.0 µg/g T-2). The chicks were housed in deep litter independent conventional system with feed and water *ad libitum* throughout the experimental study. The toxin fed chicks have exhibited a significant ($P \leq 0.05$) decrease in body weight and feed consumption. The relative weights of pancreas, thymus and bursa of Fabricius were also decreased significantly ($P \leq 0.05$). These findings were more pronounced in the combined group of AF and T-2. The data from the current study revealed that the presence of AF and T-2 in the diet can produce a synergistic affect on the performance parameters of the chicks.

Key words: Aflatoxin · T-2 Toxin · Performance · Broilers

INTRODUCTION

Cereal grains and associated by-products constitute important sources of energy for poultry. There is increasing evidence that global supplies of cereal grains for animal feedstuffs are commonly contaminated with mycotoxins [1]. Aflatoxins are secondary toxic metabolites produced by certain strains of fungi, e.g. *Aspergillus flavus* a nd *Aspergillus parasiticus* species. Aflatoxin B1 (AFB1), the most toxic of all aflatoxins (AFB1, AFB2, AFG1 and AFG2), is produced by certain strains of fungi in greater quantities than in others [1]. In poultry, aflatoxin ingestion leads to "Aflatoxicosis" syndrome which is characterized by retardation of growth, feed consumption, feed conversion efficiency and bruising, immunosuppression and mortality [1].

The T-2 is a highly toxic type A trichothecene mycotoxin produced by different *Fusarium* species, mainly *F. sporotrichoides* and to a lesser extent by *F. poae*. Both AF and T-2 are important to the poultry industry due to their synergistic toxicity and occurrence in the feeds. Co-contamination of cereal grains with mycotoxins produced by different fungal genera, including *Fusarium* and *Aspergillus* has been reported to increase the toxicity symptoms in poultry [2, 3]. Broilers fed diets containing 4ppm T-2 and 2.5ppm AF showed synergistic effect between T-2 and AF [2]. Both of these mycotoxins in combination produce a significant interaction effect on body weight. Additives effects of dietary T-2 and AF are also observed in broilers receiving 8ppm T-2 and 3.5ppm AF [4]. Combination of both the toxins decreases the body weight gain to a greater level than did either of the toxins. Synergistic toxic effects between T-2 (4ppm) and AF (2.5ppm) on relative weights of kidney, gizzard and heart is also reported, where the weights of these organs increased more than those recorded in the groups, received either of the toxin. Increased relative weights of liver, kidney, proventriculus, gizzard, spleen and pancreas were seen in broilers by feeding AF and T-2 combination [4]. Fraga *et al.* [5] reported significant interaction of AF (0.3ppm) and T-2 (3ppm) for their additive effects on body weight and feed intake. Therefore, the aim of this study was initiated to characterize the interaction between AF and T-2 in young broiler chickens at lower levels.

Corresponding Author: M. Manafi, Department of Animal Science, Malayer University, Malayer, Iran.

MATERIALS AND METHODS

Experimental Animals and Design: One hundred and sixty eight, unsexed one-day old commercial Cobb broiler chicks were wing banded, weighed and assigned to a 2X2 factorial arrangement with control (0.0), two levels of AF (0.0 and 0.5ppm), two levels of T-2 (0.0 and 2.0ppm) and combination of 0.5ppm AF +2.0ppm T-2 (AF+T-2) in a Completely Randomized Design manner, forming a total of 4 dietary treatments with three replicates and fourteen chicks per replicate in each group.

Experimental Housing, Management and Test Diet: Each replicate group of chicks was housed in an independent pen, conventional sided deep litter house. Chicks in all the replicates were reared up to five weeks of age under uniform standard conditions throughout the study. Brooding was done till three weeks of age using incandescent bulbs. Each pen was fitted with an automatic bell type drinker and a hanging tubular feeder. Chicks were provided continuous light throughout the study. AF and T-2 were produced using the pure culture of *Aspergillus parasiticus* MTCC 1894 and *Fusarium sporotrichoides* MTCC 1894 (Source: Microbial Type Culture Collection and Gene Bank, IMT, Chandigarh, 160 036, India) grown on potato dextrose agar. Then AF and T-2 toxins produced on relevant Medias were extracted as described by Rukmini and Bhat [6] and quantified by thin layer chromatography (TLC) [7].

Compounded feed was analyzed for the presence of AF and T-2 before including the rice and wheat culture materials, then the diets were prepared by incorporating required quantities of rice/wheat culture powder containing AF/T-2 into the diet so as to give the levels of 0.5ppm of AF B_1 and 2.0ppm of T-2. The given toxin levels were finally cross-checked by TLC method of analysis.

Basal diet was formulated and compounded to meet the nutrient requirements of commercial broilers during the starter (0-3 wks) (2895 Kcal/kg ME and 20.84% CP) and finisher (4-5 wks) (2994 Kcal/kg ME and 18.58% CP) feed. Chicks were provided *ad libitum* feed and water throughout the study. Feeding of test diets was commenced at first day of age and continued till the termination of experiment at five weeks of age. Chicks were vaccinated against Newcastle Disease (ND) on 7^{th} day using F_1 strain (Ventri's Biologicals, Bangalore, India) and against Infectious Bursal Disease (IBD) on 14^{th} day using intermediate strain (Ventri's Biologicals, Bangalore, India) [8]. Both the vaccines were given by oculonasal and ocular routes.

Data Collection: At the end of the trial, body weight and feed consumption were recorded and gain in weight and feed efficiency were calculated. Six birds from each replicate were sacrificed by cutting the jugular vein at the end of the trial and the weights of internal organs such as liver, kidney, gizzard, pancreas, spleen, thymus and bursa were recorded and expressed as grams per kilogram live body weight. The experimental data were analyzed statistically by using the General Linear Model procedure of Statistical Analysis System (SAS®) software [9] Revise Ref. S. No. and following references accordingly]. Duncan multiple range test was employed for comparison of the means [10]. The results of this study were subjected to one way ANOVA test.

RESULTS AND DISCUSSION

Dietary AF and T-2 produced lower body weight, feed consumption and higher feed conversion ratio significantly ($P \leq 0.05$) at the end of fifth week, when compared to controls (Table 1). In the combination group of AF+T-2, further suppression of body weight and feed consumption has been noticed during the experimental period. All the broilers were examined for oral lesions. However, oral lesions were found only in birds given the T-2. Growth depression and decreased feed consumption were recorded consistently in AF and T-2 toxicities by different scientists [11, 12], which are in agreement with the findings of the present study. The growth depression could be attributed to the AF inhibitory action on protein synthesis as well as poor nutrient utilization [13]. The T-2 is extremely caustic and produces radiomimetic action on dividing cells of organs like intestine [14]. The toxin also causes inhibition of protein synthesis by binding to ribosomes [4, 14]. This property might be possibly responsible for growth depression and poor feed conversation in T-2 toxicity. However, poor feed efficiency in AF and T-2 toxicity was consistently observed by many researchers [12, 13].

The relative weights of various organs expressed as grams per kilogram live body weight were presented in Table 2. AF showed significant ($P \leq 0.05$) increase in the size of liver, kidney, gizzard and spleen and decreased weight of pancreas, bursa of fabricius and thymus. Inclusion of T-2 in the diet showed significant ($P \leq 0.05$) increase in the relative weights of kidney and spleen and decreased weight of pancreas, bursa of fabricius and thymus. In the combination treatment, significant ($P \leq 0.05$) increase in relative weights of liver, kidney, gizzard, spleen and decreased weight of pancreas, bursa of fabricius and thymus.

Table 1: Effects of aflatoxin and T-2 toxin on body weight, feed consumption and feed conversion ratio of broilers at the fifth week.

Aflatoxin (µg/g)	T-2 Toxin (µg/g)	Body weight (g)	Feed consumption (g/bird)	Feed conversion ratio
0	0.0	2513±6.07[a]	4799.8±6.07[a]	1.91±0.003[a]
0.5	0.0	2306±2.92[b]	4819.5±2.92[b]	2.09±0.005[c]
0	2.0	2282.0±3.70[c]	4586.8±15.56[c]	2.01±0.00[b]
0.5	2.0	949.37±4.48[d]	4421.5±7.24[d]	2.16±0.01[d]

[a-f] Means in column with different superscripts differed significantly at (P≤0.05)

AF: 0.5ppm and T-2: 2ppm

Table 2: Effects of aflatoxin and T-2 toxin on relative weights of organs (grams per kg body weight) in broilers

Aflatoxin (µg/g)	T-2 Toxin (µg/g)	Liver	Kidney	Gizzard	Pancreas	Spleen	Bursa	Thymus
0	0.0	27.60±0.76[c]	8.16±0.16[a]	24.53±0.60[a]	5.10±0.17[b]	1.57±0.28[b]	1.69±0.02[a]	4.36±0.21[a]
0.5	0.0	33.00±0.57[a]	9.66±0.16[b]	25.67±1.20[b]	4.66±0.33[a]	1.66±0.16[a]	1.16±0.16[b]	2.66±0.33[b]
0	2.0	28.00±0.58[cd]	8.67±0.33[bc]	24.33±0.67[abc]	4.67±0.33[a]	1.83±0.17[a]	1.33±0.17[a]	3.33±0.33[bc]
0.5	2.0	32.33±0.88[a]	10.67±0.33[d]	25.33±0.33[bc]	4.33±0.33[a]	1.83±0.17[a]	1.00±0.29[c]	2.33±0.33[b]

[a-d] Means in column with different superscripts differed significantly at (p≤0.05)

AF: 0.5ppm and T-2: 2ppm

AF showed significant (P≤0.05) increase in the size of liver, kidney, gizzard and spleen and decrease weight of pancreas, bursa of fabricius and thymus which is in agreement Shaline *et al.* [16]. Addition of T-2 toxin in the diet showed significant (P≤0.05) increase in the relative weights of the kidney and spleen and decreased weight of pancreas, bursa of fabricius and thymus. The increase in liver and kidney weights are in accordance with the findings of many investigators [17- 20].

The increase in the weights of liver and kidney signifies the accumulation of lipid in these organs because fat metabolism primarily occurs in the liver, while lipidemia with subsequent fat deposition might contribute for increased kidney weights as well as severe inflammation [4, 21]. The increase in organ weights in AF+T-2 was reported [22]. Thaxton *et al.* [23] opined that the increased weights could be attributed to increased lipid deposition in the liver due to impaired fat metabolism which brings appreciable changes in the general functioning and gross appearance of liver [23]. The effects on gizzard are believed to be as a result of severe inflammation and the resultant thickening of the mucosa [4, 23, 24].

In the present experimental study, both AF and T-2 produced deleterious effects on the performance, decrease in body weight and feed consumption of the birds. The relative weights of pancreas, thymus and bursa of Fabricius were also reduced. The abnormalities are more pronounced in the combined toxicity of AF and T-2. Based on these findings, it can be concluded that AF and T-2 act synergistically at low level and hamper the production in the birds.

REFERENCES

1. Hagler, W.M., K. Tyczkowska and P.B. Hamilton, 1984. Simultaneous occurrence of deoxynivalenol, zearalenone and aflatoxin in 1982 scabby wheat from Midwestern United States. Appl. Environmen. Microbiol., 47: 151-154.
2. Huff, W.E., R.B. Harvey, L.F. Kubena and G.E. Rottinghaus, 1988. Toxic synergism between aflatoxin and T-2 toxin in broiler chickens. Poult. Sci., 67: 1418-1423.
3. Manafi, M., K. Mohan and M. Noor Ali, 2011. Effect of Ochratoxin A on Coccidiosis-Challenged Broiler Chicks. World Mycotoxin J., 4: 177-181.
4. Kubena, L.F., R.B. Harvey, W.E. Huff, D.E. Carier, T.D. Phillips and G.E. Rottinghaus, 1990. Efficacy of hydrated sodium calcium aluminosilicate to reduce the toxicity of aflatoxin and T-2 Toxin. Poult. Sci., 69: 1078-1086.
5. Fraga, M.E., F. Curvello, M.J. Gatti, L.R. Cavaglieri, A.M. Dalcero and C.A. Darocha Rosa, 2007. Protemtial aflatoxin and ochratoxin A production by *Aspergillus* species in poultry feed processing. Vet. Res. Communications, 31: 345-353.
6. Rukmini, C. and R.V. Bhat, 1978. Occurrence of T-2 toxin in Fusarium infested sorghum from India. J. Agri. Food Chemist., 26: 647-649.
7. A.O.A.C., 1995. Official Methods of Analysis. 16th Ed., Association of Official Analytical Chemists, Washington, D.C.
8. Calnek, B.W., H.J. Barnes, C.W. Beard, W.M. Reid and Jr. H.W. Yoder, 1992. Diseases of Poultry, 9th ed., Wolfe Publishing, Ltd., USA.

9. S.A.S., 2000. Statistical Analysis Systems User's Guide: Statistics. SAS Institute Inc., Cary, NC, USA.
10. Duncan, D.B., 1955. Multiple range and multiple F tests. Biometrics, 11: 1-42.
11. Raju, M.V.L.N. and G. Devegowda, 2000. Influence of esterified glucomannan on performance and organic morphology, serum biochemistry and hematology in broilers exposed to individual and combined mycotoxicosis (aflatoxin, ochratoxin and T-2 toxin). British Poult. Sci., 41: 640-650.
12. Arvind, K.L., V.S. Patil, G. Devegowda, B. Umakantha and S.P. Ganpule, 2003. Efficacy of Esterified glucomannan to counteract Mycotoxicosis in naturally contaminated feed on performance, serum biochemical and hematological parameters in broilers. Poult. Sci., 82: 571-576.
13. Marquardt, D.R. and A. Frohlich, 1992. A review of recent advances in understanding ochratoxicosis. J. Anim. Sci., 70: 3968-3988.
14. Ueno, Y., 1997. Mode of action of trichothecenes. Pure and Applied Chemistry, 49: 1737-3745.
15. Ahmadi, M., 2010. Effect of Turmeric (*Curcumin longa*) Powder on Performance, Oxidative Strees State and Some of Blood Parameters in Broilers Fed on Diet Containing Aflatoxin B1. Global Veterinaria, 5: 312-317.
16. Shaline, K.S.B., B. Howrth, Jr. and R.D. Wyatt, 1980. Effect of dietary aflatoxin in reproductive performance on mature white leghorn. Poult. Sci., 59: 1311-1315.
17. Girish, C.K. and G. Devegowda, 2004. Evaluation of modifies glucomannan (Mycosorb) and HSCAS to ameliorate the individual and combined toxicity of aflatoxin and T-2 toxin in broiler chickens. Aust. Poult. Sci. Symp. Sydney, Australia, 16: 126-129.
18. Arvind, K.L., V.S. Patil, G. Devegowda, B. Umakantha and S.P Ganpule, 2003. Efficacy of Esterified glucomannan to counteract Mycotoxicosis in naturally contaminated feed on performance, serum biochemical and hematological parameters in broilers. Poult. Sci., 82: 571-576.
19. Ghahri, H., R. Habibian and M. Abdolah Fam, 2010. Effect of Sodium Bentonite, Mannan Oligosaccharide and Humate on Performance and Serum Biochemical Parameters During Aflatoxicosis in Broiler Chickens. Global Veterinaria, 5: 129-134.
20. Salahi, A., S.N. Mousavi, F. Foroudi, M.M. Khabisi and M. Norozi, 2011. Effects of in ovo Injection of Butyric Acid on Broiler Breeder Eggs on Hatching Parameters, Chick Quality and Performance. Global Veterinaria, 7: 468-477.
21. Miazzo, R., M.F. Pevalta, C. Magnoli, M. Salvano, S. Ferrero, S.M. Chiacchiera, E.C.Q. Carralno, C.A.R. Rosa and A. Dalcero, 2005. Efficacy of sodium bentonite as a detoxifier of broiler fed contaminated with aflatoxin and fumonisin. Poult. Sci., 84: 1-8.
22. Verma, J., T.S. Johri, B.K. Swan and S. Ameena, 2004. Effect of graded levels of aflatoxin and their combination on the performance and immune response of broilers. British Poult. Sci., 45: 512-518.
23. Thaxton, J.P., J.T. Timb and B. Jaotpm, 1974. Immunosuppression in chicken by aflatoxin. Poult. Sci., 53: 721-725.
24. Azarakhsh, Y., A. Sabokbar and M. Bayat, 2011. Incidence of the Most Common Toxigenic Aspergillus Species in Broiler Feeds in Kermanshah Province, West of Iran. Global Veterinaria, 6: 73-77.